

- CS
- 5
80. Birnboim et al., Nucleic Acids Research 7, 1513 (1979).
81. Maxam and Gilbert, Proc. Nat. Acad. Sci. 74, 560 (1977).
82. McGrath and Levinson, Nature 295, 423 (1982).
83. Itakura et al., Science 198, 1056 (1977).
84. Crea et al., Proc. Natl. Acad. Sci. 75, 5765 (1978).--

---

IN THE CLAIMS:

Please cancel claims 1, 6, 9-17, and 24-25, without prejudice.

Please amend the remaining claims as follows:

---

CS

2. (Amended) A process for obtaining a mature protein heterologous to yeast as a product of yeast expression, which process comprises:

(a) transforming a yeast organism with an expression vehicle comprising [the] a DNA sequence encoding [substantially the] an Arg C-terminal pre-pro peptide of yeast alpha factor operably connected in translation reading frame without intervening Glu (or Asp)-Ala dipeptide repeats to a DNA sequence encoding a mature protein heterologous to the yeast organism;

(b) culturing the transformed organism; and

(c) recovering the mature protein from the culture.

3. (Amended) A process for obtaining a mature protein heterologous to yeast as a product of yeast expression, processing and secretion, which process comprises:

(a) transforming a yeast organism with an expression vehicle comprising [the DNA sequence of the promoter] yeast alpha factor promoter DNA sequence operably linked to [substantially the] an Arg C-terminal pre-pro peptide sequence of [the] yeast alpha factor

09 [gene] which is operably connected in translation reading frame without intervening Glu (or Asp)-Ala dipeptide repeats to a DNA sequence encoding a mature protein heterologous to the yeast organism;

(b) culturing the transformed yeast organism; and

(c) recovering the mature protein from its supporting medium.

4. (Amended) A process for secreting a mature protein heterologous to yeast into the supporting medium, which process comprises:

(a) transforming a yeast organism with an expression vehicle comprising [the] a DNA sequence encoding [substantially the] an Arg C-terminal pre-pro peptide of yeast alpha factor, operably connected in translation reading frame without intervening Glu (or Asp)-Ala dipeptide repeats to a DNA sequence encoding a mature protein heterologous to the yeast organism; and

(b) culturing the transformed organism.

5. (Amended) The process of Claim 4 wherein said DNA [sequences are] sequence encoding an Arg C-terminal pre-pro peptide of yeast alpha factor is under the control of alpha factor promoter.

010 7. (Amended) [The] An expression vehicle [of Claim 6 which also includes the] comprising yeast alpha factor promoter DNA sequence operably connected to a DNA sequence encoding a mature protein heterologous to the yeast organism, and also comprising a DNA sequence encoding [substantially the] an Arg C-terminal pre-pro peptide of yeast alpha factor operably linked in translation reading frame without intervening Glu (or Asp)-Ala dipeptide repeats upstream to the DNA sequence encoding [a] the mature protein heterologous to the yeast organism, wherein the protein is in discrete form unaccompanied by any substantial peptide

*C10*  
presequence or other artifact of expression, as a product of yeast expression, processing and secretion.

8. (Amended) A yeast expression vehicle comprising [the] a DNA sequence encoding [substantially the] an Arg C-terminal pre-pro peptide of yeast alpha factor [gene] operably connected in translation reading frame without intervening Glu (or Asp)-Ala dipeptide repeats to a DNA sequence encoding a mature protein heterologous to the yeast organism, wherein the protein is in discrete form unaccompanied by any substantial peptide presequence or other artifact of expression, as a product of yeast expression, processing and secretion.

*OK*  
18. (Amended) The expression vehicle of Claim 7 wherein the DNA sequence encoding [heterologous] the mature protein heterologous to the yeast organism encodes [for a protein selected from the group consisting of] human interferon [, bovine interferon, tissue plasminogen activator, and rennin].

19. (Amended) The expression vehicle of Claim 8 wherein the DNA sequence encoding [heterologous] the mature protein heterologous to the yeast organism encodes [for a protein selected from the group consisting of] human interferon [, bovine interferon, tissue plasminogen activator, and rennin].

In claim 22, line 1, change "transformed with" to  
--comprising--.

In claim 23, line 1, change "transformed with" to  
--comprising--.

Please add new claims as follows:

CI2  
--26. The expression vehicle of Claim 7 wherein the DNA sequence encoding the mature protein heterologous to the yeast organism encodes bovine interferon.--

--27. The expression vehicle of Claim 7 wherein the DNA sequence encoding the mature protein heterologous to the yeast organism encodes tissue plasminogen activator.--

--28. The expression vehicle of Claim 7 wherein the DNA sequence encoding the mature protein heterologous to the yeast organism encodes rennin.--

--29. The expression vehicle of Claim 8 wherein the DNA sequence encoding the mature protein heterologous to the yeast organism encodes bovine interferon.--

--30. The expression vehicle of Claim 8 wherein the DNA sequence encoding the mature protein heterologous to the yeast organism encodes tissue plasminogen activator.--

--31. The expression vehicle of Claim 8 wherein the DNA sequence encoding the mature protein heterologous to the yeast organism encodes rennin.--

--32. A DNA molecule comprising a DNA sequence encoding an Arg C-terminal pre-pro peptide of yeast alpha factor operably connected in translation reading frame without intervening Glu (or Asp)-Ala dipeptide repeats to a DNA sequence encoding a mature protein heterologous to the yeast organism.--

--33. The DNA molecule of claim 32 wherein the DNA sequences are under the control of alpha factor promoter.--

--34. The DNA molecule of claim 32 wherein the DNA sequence encoding the mature protein heterologous to the yeast organism encodes insulin-like growth factor.--

--35. The DNA molecule of claim 32 wherein the DNA sequence encoding the mature protein heterologous to the yeast organism encodes human interferon.--

--36. The DNA molecule of claim 32 wherein the DNA sequence encoding the mature protein heterologous to the yeast organism encodes bovine interferon.--

--37. The DNA molecule of claim 32 wherein the DNA sequence encoding the mature protein heterologous to the yeast organism encodes tissue plasminogen activator.--

--38. The DNA molecule of claim 32 wherein the DNA sequence encoding the mature protein heterologous to the yeast organism encodes rennin.--

--39. The process of claim 2 wherein the DNA encoding all of the Glu (or Asp)-Ala dipeptide repeats has been deleted from the pre-pro peptide of the yeast alpha factor DNA--.

--40. A process for obtaining a mature protein heterologous to yeast as a product of yeast expression, which process comprises:

(a) culturing a yeast organism comprising an expression vehicle comprising a DNA sequence encoding an Arg C-terminal pre-pro peptide of yeast alpha factor operably connected in translation reading frame without intervening Glu (or Asp)-Ala dipeptide repeats to a DNA sequence encoding a mature protein heterologous to the yeast organism; and

(b) recovering the mature protein from the culture.--

--41. A process for obtaining a mature protein heterologous to yeast as a product of yeast expression, processing and secretion, which process comprises:

(a) culturing a yeast organism comprising an expression vehicle comprising yeast alpha factor promoter DNA sequence operably linked to an Arg C-terminal pre-pro peptide sequence of yeast alpha factor which is operably connected in translation reading frame without intervening Glu (or Asp)-Ala dipeptide repeats to a DNA sequence encoding a mature protein heterologous to the yeast organism; and

C12 (b) recovering the mature protein from its supporting medium.--

--42. A process for secreting a mature protein heterologous to yeast into the supporting medium, which process comprises culturing a yeast organism comprising an expression vehicle comprising a DNA sequence encoding an Arg C-terminal pre-pro peptide of yeast alpha factor, operably connected in translation reading frame without intervening Glu (or Asp)-Ala dipeptide repeats to a DNA sequence encoding a mature protein heterologous to the yeast organism.--

--43. The process of claim 40 wherein said DNA sequence encoding an Arg C-terminal pre-pro peptide of yeast alpha factor is under the control of alpha factor promoter.--

--44. The process of claim 42 wherein said DNA sequence encoding an Arg C-terminal pre-pro peptide of yeast alpha factor is under the control of alpha factor promoter.--

--45. The process of claim 2 wherein said DNA sequence encoding an Arg C-terminal pre-pro peptide of yeast alpha factor is under the control of alpha factor promoter.--